## Microbial Flora of Pecan Meat<sup>1</sup>

## J. R. CHIPLEY<sup>2</sup> AND E. K. HEATON

Department of Food Science, University of Georgia College of Agriculture, Georgia Experiment Station, Experiment, Georgia 30212

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Microorganisms found associated with commercially shelled pecan meats are numerous and varied. No bacteria and only two fungal microorganisms, *Aspergillus clavatus* and *Tricothecium* species, were found on aseptically shelled pecans.

Bacteria of the coliform group, especially Escherichia coli, are commonly used as indicators of fecal pollution. Ostrolenk and Hunter (8), Ostrolenk and Welch (9), Kokal (5), and other workers have reported that nut meats in unbroken shells do not contain this bacterium and that its presence in shelled nut meats reflects unsanitary processing practices. Other bacteria, including Bacillus and Streptococcus, have been identified on several types of nut meats used in ice cream mixes (11). Fungi, especially aflatoxigenic isolates of Aspergillus flavus, have been isolated from pecans by Lillard et al. (7). However, there

face-sterilized by total submersion for 2 min in a solution of  $1.0 \text{ mm HgCl}_2$  and then rinsed with sterile distilled water. The meats were then aseptically removed and placed in sterile petri dishes. For bacterial isolation, sterilized Trypticase soy agar was then poured over these samples and over samples of commercially shelled pecans in other dishes, and the dishes were gently rotated and allowed to solidify. For fungal isolation, samples were plated in 2% (w/v) agar (Difco) in the same manner. Bacterial samples were incubated at 37 C; fungal samples were incubated at 22 C.

Isolation and identification of all bacteria were

Table 1. Identification of microorganisms found in pecan meats

Bacteria	Per cent <sup>a</sup>	Fungi	Per cent
Pseudomonas effusa	10	Penicillium notatum	30
Corynebacterium paurometabolum	10	Aspergillus clavatus	$15^{b}$
Escherichia coli	20	A. niger	15
Leuconostoc mesenteroides	20	Fusidium species	15
Proteus vulgaris	10	Tricothecium species	15c
Aerobacter aerogenes	30	•	
Clostridium species	10		

<sup>&</sup>lt;sup>a</sup> Percentage of commercially shelled samples tested that were positive for indicated microorganism.

appears to have been no attempt to isolate and identify all of the microflora present on a particular sample of nut meat. It was the purpose of this investigation to isolate and identify all of the bacteria and fungi present on samples of both shelled and unshelled pecan meats.

Shelled and unshelled pecan meat samples were obtained from two commercial operations and stored at 4 C until microbiological examination could be conducted. Unshelled pecans were sur-

conducted in accordance with published procedures (2, 3, 10; *Bergey's Manual*, 7th ed.) Petri dishes containing fungi were incubated until sporulation of the colonies occurred (approximately 2 weeks). Then, isolation and identification of colonies were conducted in accordance with the *Laboratory Manual for Medical Mycology* (1).

Thin sections of both commercially shelled and aseptically shelled pecan meats were examined for the presence of fungal hyphae and were prepared by impregnation of meats with liquefied paraffin wax for 48 hr at 80 C. Samples were then cooled, and thin sections having a thickness of 25  $\mu$ m were prepared by using a Spencer rotary microtome.

<sup>&</sup>lt;sup>b</sup> Per cent isolated from aseptically shelled pecans was 4.

<sup>&</sup>lt;sup>c</sup> Per cent isolated from aseptically shelled pecans was 5.

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<sup>&</sup>lt;sup>2</sup> Present address: Department of Poultry Science, Ohio State University, Columbus, Ohio 43216.

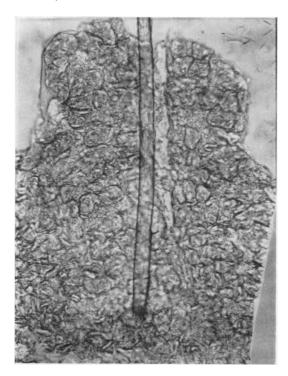


Fig. 1. Thin-section micrograph of commercially shelled pecan meat showing penetration of tissue by hypha of fungus.  $\times$  200.

The results of this study indicate that the microorganisms found associated with commercially shelled pecan meats are numerous and varied (Table 1). No bacteria and only two fungal microorganisms. A. clavatus and Tricothecium species, were found on aseptically shelled pecans. In commercial operations, the mechanical processes of washing, cracking, separating, and sorting out the nut meats apparently serve to inoculate them as well. Other authors (4, 6, 8) have indicated that this might be the case.

Fungal spores can apparently germinate and produce actively growing hyphal tissue capable of penetrating the pecan tissue (Fig. 1). Although isolations of two fungi were made from aseptically

shelled samples held under recommended conditions of storage, no evidence of hyphal growth or penetration of tissue could be obtained.

Hyndman (4) stated that other pathogenic microorganisms associated with the presence of *E. coli* introduce a health factor which is important to regulatory agenicies concerned with consumer protection. The present study indicates that unless pecan meats are stored under conditions of low temperature and humidity immediately after harvesting and processing, problems involving the safety of this product for human consumption could be encountered.

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